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A study of preoperative methionine-depleting parenteral nutrition plus chemotherapy in gastric cancer patients

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Abstract

AIM To investigate the interference of methionine-free paren teral nutrition plus 5-Fu (MetTPN+5-Fu) in gastric cancer cell kinetics and the side effects of the regimen.

METHODS Fifteen patients with advanced gastric cancer were randomly divided into two groups, 7 patients were given preoperatively a seven-day course of standard parenteral nutrition in combination with a five-day course of chemotherapy (sTPN+5-Fu), while the other 8 patients were given methionine-deprived parenteral nutrition and 5-Fu (MetTPN+5-Fu). Cell cycles of gastric cancer and normal mucosa were studied by flow cytometry (FCM). Blood samples were taken to measure the serum protein, methionine (Met) and cysteine (Cys) levels, and liver and kidney functions.

RESULTS As compared with the results obtained before the treatment, the percentage of G_0/G_1 tumor cells increased and that of S phase decreased in the MetTPN+5-Fu group, while the contrary was observed in the sTPN+5-Fu group. Except that the ALT, AST and AKP levels were slightly increased in a few cases receiving -MetTPN+5-Fu, all the other biochemical parameters were within normal limits. Serum Cys level decreased slightly after the treatment in both groups. Serum Met level of patients receiving sTPN+5-Fu was somewhat higher after treatment than that before treatment; however, no significant change occurred in the MetTPN+5-Fu group, nor operative complications in both groups.

CONCLUSION -MetTPN+5-Fu exerted a suppressive effect on cancer cell proliferation, probably through a double mechanism of creating a state of "Met starvation" adverse to the tumor cell cycle, and by allowing 5-Fu to kill specifically cells in S phase. Preoperative short-term administration of -MetTPN+5-Fu had little undesirable effect on host metabolism.

INTRODUCTION

Methionine (Met) has been reported to be an essential amino acid in tumor cell metabolism. Recent researches have demonstrated that proliferation was inhibited in some tumor cell lines when Met in the culture medium was replaced by its precursor homocysteine (Hcy), whereas normal cells grew well as controls; and once regaining Met, tumor cells recovered their usual activity of proliferation; these characteristics of tumor cells were referred to as Met dependency[1,2]. In the Met depleted environment, the metabolism of nucleic acid, protein and biological membrane of the tumor cells were disturbed and their cell cycle deranged. If Met starvation was combined with the application of phase-specific chemotherapeutical agents, it would prohibit the proliferation and metastasis of tumor cells. On the basis of our previous experiments, further studies are designed to elucidate the influence of preoperative Met deprived parenteral nutrition plus 5-Fu on the cell cycle in the advanced gastric cancer (AGC) patients, and to explore whether any adverse effect to host metabolism could be produced, by monitoring the serum Met and cysteine (Cys) levels and blood biochemistry parameters.

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PATIENTS AND METHODS

Patients

From June to December 1996, fifteen AGC patients (8 males and 7 females) diagn osed by endoscopy and pathology, entered into the study. Their average age was 55 (39-71) years and the average body weight was 55 (33-78) kg. All the patients had normal liver and kidney functions and were free from metabolic diseases.

Groups

The patients were randomized to 2 groups, 7 patients of the control group were administered Met-containing standard parenteral nutrition plus chemotherapy (sTPN+5-Fu), and 8 of the study group were given Met-deprived parenteral nutrition plus chemotherapy (MetTPN+5-Fu).

Regimens of nutritional support and chemotherapy

All patients were permitted to take a small amount of soup and water, no blood products were used. The non-protein energy in the TPN was supplied by glucose and fat emulsion (20% Intralipid, SSPC). The amino acid solution used in the control group was HBC11S (Amino Acid Company, Tianjin) while that used in the study group was prepared cooperatively by Rui Jin Hospital and Shanghai Chang Zheng Pharmaceutical Plant (No. S 95-001 Rui Jin, Shanghai, China). The amount of fat-soluble and water-soluble vitamins, trace elements and electrolytes was same for both groups. The mean non-protein energy was 125 kJ/(kg·d) for both groups; the nitrogen intake was 0.165g/(kg·d) and 0.163g/(kg·d) respectively for the control and the study groups. The TPN was administered from d1 to d7, and 500mg/d of 5-Fu from d2 to d6. Operation was carried out on d8.

Laboratory tests

Cell kinetics Tissue samples of gastric cancer and normal ga stric mucosa obtained through endoscopy at the beginning of the study and during gastrectomy were managed mechanically to produce monocellular suspension, and were then submitted to flow cytometry (FCM) study for cell kinetics.

Blood biochemistry Blood samples were collected before and after the treatment for biochemical studies: liver function tests: ALT, AST, γ-GT and AKP; kidney function tests: BUN and Cr; serum proteins: total protein (TP), albumin (Alb), transferrin (Tf) and prealbumin (PA); and serum Met and Cys.

Statistical analysis

Paired *t* test and Student *t* test were used.

RESULTS

Cell cycle study

In 2 patients of the control group, the number of tissue samples was incomplete because gastrectomy was not performed eventually, resulting in a total of 13 patients who entered the FCM analysis. In the study group, compared to the pre treatment data, the percentage of tumor cells in G_0/G_1 phases increased after the treatment, while the percentage of S phase cells decreased; there was no obvious change in the percentage of 2/M cells, but the percentage of $S+G_2/M$ (representing phases of cell proliferation) decreased. In the control group, the percentage of G_0/G_1 of tumor cells decreased after the treatment while that of S phase increased. There were no obvious changes of G_2/M and $S+G_2/M$ percentage (Table 1).

Table 1 Changes in tumor cell cycle before and after treatment ($\bar{x}\pm s$)

Phase		sTPN+5-Fu (n=5)	MetTPN+5-Fu (n=8)
G_0/G_1	Before	67.10±27.68	78.30±11.01
	After	63.35±28.72	83.45±9.26
S	Before	22.22±18.60	11.96±6.41
	After	26.08±26.02	9.25±7.16
G_2/M	Before	10.66±9.87	8.49±3.89
	After	8.56±3.76	8.50±4.17
$S+G_2/M$	Before	32.88±27.68	21.21±7.18
	After	34.64±28.76	18.85±7.62

Regarding to the cell kinetics of normal mucosa, there was no significant change in both groups either before or after the treatment.

Liver and kidney function

Initially, all the patients of both groups had normal liver functions. After the treatment, blood biochemical parameters remained within normal limits in the majority of cases except that some of the parameters were slightly higher than normal in a few cases. All the patients had normal kidney functions during the study.

Serum proteins

Serum protein parameters of both groups rose after the treatment (P>0.05), except the Tf value in the control group. The increment was slightly higher in the control group than that in the study group, but there was no statistical significance (Table 2).

Table 2 Changes of serum protein levels before and after treatment ($\bar{x}\pm s$)

Serum protein	sTPN+5-Fu (n = 7)		-MetTPN+5-Fu (n = 8)	
	Before	After	Before	After
TP(g/L)	69.86±4.49	75.14±4.85	68.75±7.67	72.88±3.94
Alb(g/L)	38.89±5.95	42.29±5.22	35.57±4.98	37.85±6.09
Tf(g/L)	2.68±0.49	2.68±0.36	2.20±0.82	2.36±0.44
PA(mg/L)	225.83±44.36	312.43±96.14	181.57±49.85	219.88±70.17

Serum Met and Cys

The mean serum Met level was increased after the treatment in the c ontrol group (P=0.03738), while there was no obvious change in the study group. Serum Cys level had little changes after the treatment in both groups, with no statistical significance (Table 3).

Table 3 Changes of serum Met and Cys levels before and after treatment ($\bar{x}\pm s$)

	sTPN+5-Fu (*n*=7)		-MetTPN+5-Fu (*n*=8)	
	Before	After	Before	After
Met(mmol/L)	36.53±9.97	50.90±18.96[a]	35.26±7.05	36.64±10.27
Cys(mmol/L)	70.13±22.79	56.23±17.70	54.83±14.12	57.21±23.47

[a]$P<0.05$ *vs* before the treatment.

DISCUSSION

More than twenty years ago, Kreis *et al*[3] explored by histoculture and animal e xperimentation, the possible role of Met deprivation in the diet or TPN in suppr essing tumor growth and metastasis. By means of the FCM technique, Usami *et al*[4] studied the effect of Met-deficiency on tumor cell growth and cell kinetics and found that the environment of "Met starvation", either *in vivo* or *in vitro*, could effectively interfere with cell recycling and block the tumor cells in G_1 phase. Hoffman *et al*[5] noticed that by a method named the three-dimensional histocult ure, when Met in the culture medium was replaced by Hcy, the proliferation of most Met-dependent tumor cells was inhibited in the latter Sphase/G_2 phase. In spite of certain diversities in their experimental results, these authors shared the common view that the life cycle of tumor cells could be disturbed by "Met starvation". This concept favors the simultaneous use of phase-specific chemotherapeutic agents in an attempt to improve the overall therapeutic effects of neoplastic diseases.

Tumor tissues are composed of proliferating cells, non-proliferative and un-proliferating cell masses. The proliferating cells are those that are under going mitosis and are most sensitive to the phase-specific antitumor drugs, while those belonging to the non-proliferative or un-proliferating masses are not. By inference, more tumor cells are in the state of active proliferation, the greater effects would the phase-specific drugs exercise on the tumor cells. Maeta *et al*[6] used preoperatively the MetTPN+5-Fu therapy to a group of AGC and colorectal cancer patients, found that the inhibition rate to thymidilate synthetase in the tumor cells was significantly higher than that in the control group, the name of "biochemical modulator" was thus coined to 5-Fu by these authors. Hoshiya, Goseki, Kitamura, Taguchi and others proved the effe ctiveness in combining 5-Fu, cisplatin or adriamycin to -MetTPN in the treatment of gastric cancer and breast cancer. Kitamura et al and Taguchi et al further applied a new amino acid solution free of Met and Cys, named AO-90; this regim en was especially satisfactory in patients with distant metastasis and ascites, mounting the response rate to 45.5%[7-10].

Our previous studies[11-13] confirmed that the influence of sTPN on cell kinetics was to stimulate the tumor cells to enter into S phase; this would create an optimal condition for 5-Fu to exercise its greatest effect on tumor cells. In the present study, the effect of preoperative MetTPN+5-Fu on tumor cell kinetics in AGC patients was compared with that of sTPN+5 Fu. The results showed that, in the MetTPN+5-Fu group the percentage of tumor cells in G_0/G_1 phase increased and the percentage of cells in S and $S+G_2/M$ phases decreased. The underlying rationale was not certified whether it was due to the decline in S phase cells caused by killing effect of 5-Fu, or an improved biochemical modulator effect of 5-Fu as described by Maeta *et al*[6].

As a special therapeutical method which utilized the metabolic defect of tumor cells, there was still doubt that whether the -MetTPN+5-Fu regimen would bring about undesirable effects to the metabolism of normal cells remains to be clarified. Goseki *et al*[14] studied the nutritional status of experimental rats after administration of a Met-free amino acid solution; no obvious side e ffects were found except slight decrement of a few nutritional parameters. Kurih ara *et al*[15] used the AO-90 amino acid solution plus chemotherapy in recurrent AGC patients, who manifested only a slight increase of nausea and anorexia. In rats with experi mental gastric cancer submitted to the -MetTPN treatment, only a slight weight loss was observed as a side-effect; the serum protein levels and liver and kidney function underwent no significant change after a preoperative -MetTPN+5-Fu therapy for one week; serum Met and Cys levels were not obviously influenced; no operative complications occurred in any of the patients under study[16].

In conclusion, a short-term preoperative -MetTPN+5-Fu in AGC patients did not cause any negative effect on normal host metabolism, while producing an evident interference with gastric cancer cell kinetics.

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